EFFECTS ON BONE TISSUE IN MALE AND FEMALE TRANSGENIC MICE EXPRESSING A CONSTITUTIVELY ACTIVE ARYL HYDROCARBON RECEPTOR

Wejheden C[1], Brunnberg S[1], Ohlsson C[2], Hanberg A[1], and Lind PM[1]
[1]Institute of Environmental Medicine, Karolinska Institutet, Box 210, 171 77 Stockholm, [2]Department of Internal Medicine, Gothenburg University, Sweden

Introduction
Dioxins are ubiquitous environmental pollutants known to cause a wide spectrum of toxic effects, including reproductive and developmental effects as well as cancer [1]. Dioxins and dioxin-like compounds exert their effects by binding to the aryl hydrocarbon receptor (AhR) with high affinity. A study with the dioxin-like PCB126 shows that bone tissue composition and strength in female rat is impaired in exposed animals. Furthermore, PCB 126 has divergent effects depending on if the rats were ovariectomized or not [2]. A recent study showed that *in utero* and lactational exposure to TCDD caused alterations in the bone tissue, e.g. the length of the bones were decreased and bone mineral density was decreased [3].

Transgenic mice with a constitutively active AhR (CA-AhR) have been developed and may function as a model for studying the effects of dioxins and the mechanisms behind these. The effects in the CA-AhR mice resemble the effects of mice exposed to low doses of dioxin [4, 5]. A bone-associated protein, osteopontin, was significantly down-regulated in stomach tumors of these CA-AhR mice [6]. Osteopontin is a protein suggested to be involved in bone tissue remodelling and promotes attachment of the bone cell to the matrix, thus, changes in the expression of osteopontin may be involved in an altered bone remodelling. *In vitro* studies with an osteoblastic cell line, UMR-106, have shown that osteopontin is rapidly down regulated in these cells when exposed to TCDD [7].

Aim
The aim of this pilot study was to investigate the potential effects on bone tissue in CA-AhR mice.

Methods
The long bones of male and female mice (9-12 month old) were excised. Tibiae were analyzed with peripheral quantitative computed tomography (pQCT). Cortical parameters, including cortical bone mineral content and cortical area, were determined by mid-diaphyseal scans. Trabecular parameters, including trabecular bone mineral density, trabecular bone mineral content and trabecular bone area were determined by metaphyseal scans of the tibiae.

Results and discussion
The long bones of CA-AhR mice are significantly different compared to bones of wild type mice. In females, the cortical bone area was significantly increased while the trabecular density was decreased (Fig 2). In males, both cortical and trabecular parameters were significantly increased (Fig 1 and 3). These results therefore suggest involvement of AhR in dioxin induced bone toxicity. In conclusion the long bones of mice are affected by an active AhR. Both cortical and trabecular parameters were significantly affected in both sexes.

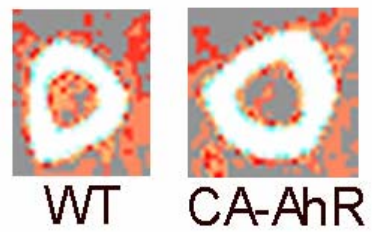

Figure 1. pQCT scans of cortical bone area in the diaphyseal part of the tibiae of male mice. The CA-AhR bone has a greater cortical area than the wild type.

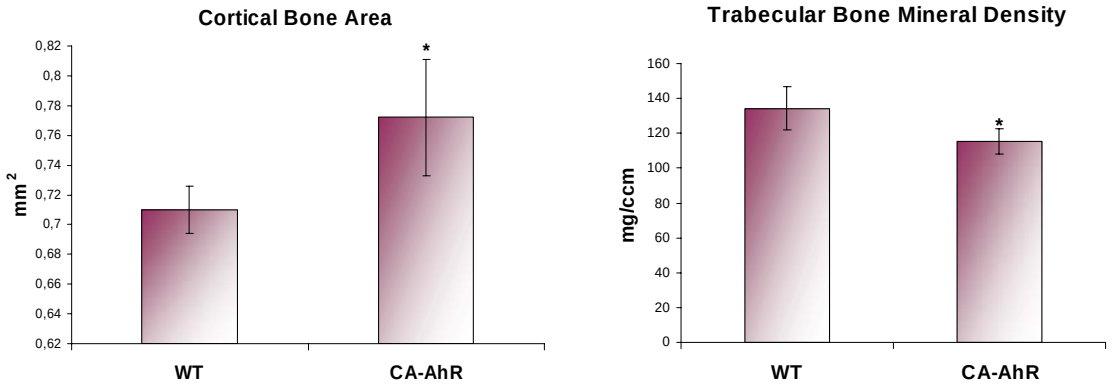

Figure 2. The cortical bone area was significantly increased in female transgenic mice, while trabecular bone mineral density was significantly decreased.

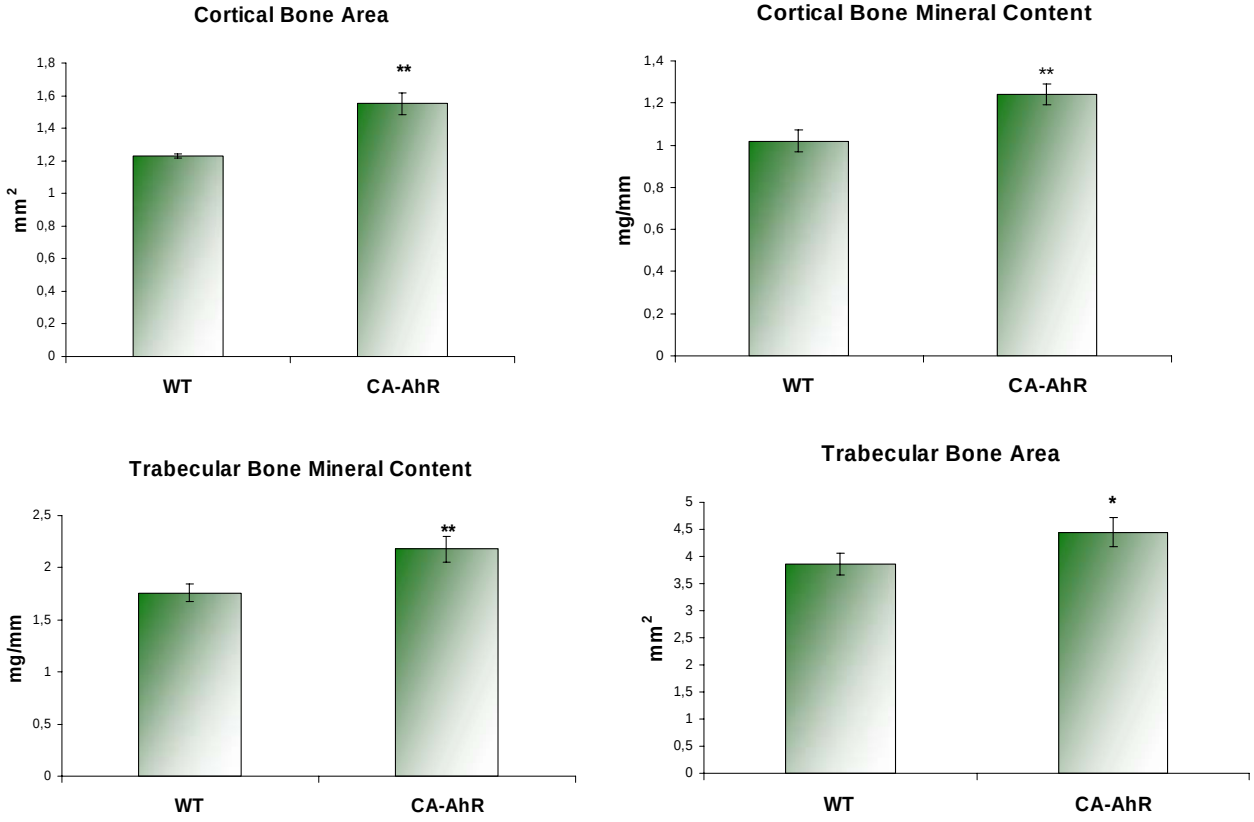

Figure 3. The cortical bone area, cortical bone mineral content, trabecular bone mineral content and trabecular bone area werv significantly increased in male transgenic mice compared to wild type.

Future studies

We are currently studying long bones from young male and female CA-AhR and wild type mice. The composition and dimension of the bones will be measured using computerized tomography specialized for bone measurements; pQCT. MicroCT will be performed to obtain detailed information of the effects on bone structure. The measurements will be performed in the central part of the diaphyses of femur and in the distal tibia. Furthermore, also specific biomarkers will be studied. Analysis of bone-related hormones and growth factors (estrogen, testosterone, insulin-like growth factor (IGF-I), PTH and 1,25-vitamin D), and biomarkers for bone resorption

(type I collagen crosslinked C-terminal telopeptide (CTX) and tartrate alkaline phosphatase (TRAP)) and formation (osteocalcin and alkalaine phosphatase (ALP)) will be analyzed.

References

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